(11) EP 2 395 014 A3

(12)

EUROPEAN PATENT APPLICATION

(88) Date of publication A3: 11.07.2012 Bulletin 2012/28

(43) Date of publication A2: **14.12.2011 Bulletin 2011/50**

(21) Application number: 11166330.8

(22) Date of filing: 04.10.2006

(51) Int CI.:

C07K 14/435 (2006.01) C07H 21/04 (2006.01) A61K 47/42 (2006.01) C07K 19/00 (2006.01) C12N 15/63 (2006.01)

(84) Designated Contracting States:

AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HU IE IS IT LI LT LU LV MC NL PL PT RO SE SI SK TR

Designated Extension States:

AL BA HR MK RS

(30) Priority: 05.10.2005 US 723766 P

(62) Document number(s) of the earlier application(s) in accordance with Art. 76 EPC: 06790323.7 / 1 931 702

(71) Applicant: Commonwealth Scientific and Industrial
Research Organization
Campbell, Australian Capital Territory 2612 (AU)

(72) Inventors:

Sutherland, Tara, D.
 Watson, Australian Capital Territory 260 (AU)

Haritos, Victoria, S.
 O'connor, Australian Capital Territory 2 (AU)

 Trueman, Holly Frenchs Forest, New South Wales 2086 (AU)

 Sriskantha, Alagacone Nicholls, Australian Capital Territory 2 (AU)

 Weisman, Sarah Griffith, Australian Capital Territory 2 (AU)

Campbell, Peter, M.
 Cook, Australian Capital Territory 2614 (AU)

(74) Representative: Harrison, Susan Joan Mewburn Ellis LLP 33 Gutter Lane London EC2V 8AS (GB)

(54) Silk proteins

(57) The present invention provides silk proteins, as well as nucleic acids encoding these proteins. The present invention also provides recombinant cells and/or organisms which synthesize silk proteins. Silk proteins

of the invention can be used for a variety of purposes such as in the manufacture of personal care products, plastics, textiles and biomedical products.



Application Number

EP 11 16 6330

Category	Citation of document with in of relevant passa	dication, where appropriate, ges		Relevant o claim	CLASSIFICATION OF THE APPLICATION (IPC)
X		o05-02-14), estris larval caste ris cDNA clone QL1 39, accession no.	1- 15	5,13,	INV. C07K14/435 C07K19/00 C07H21/04 C12N15/63 A61K47/42
X	DATABASE EMBL [Onli 27 August 2005 (200 "JGI_CABJ9548.fwd N tropicalis cDNA clo mRNA sequence.", XP002661622, retrieved from EBI EM_EST:DT432295 Database accession * sequence *	5-08-27), IH_XGC_tropSki1 Xenopu ne IMAGE:7874366 5', accession no.	15	5,13,	TECHNICAL FIELDS SEARCHED (IPC)
X	DATABASE UniProt [0 13 September 2005 ("SubName: Full=Muci protein (MASP), put XP002661623, retrieved from EBI UNIPROT:Q4DYV1 Database accession * sequence *	2005-09-13), n-associated surface ative;", accession no. no. Q4DYV1		5,7, ,13,15	
	Place of search	Date of completion of the search			Examiner
	Munich	11 May 2012		Cha	vanne, Franz
X : parti Y : parti docu	ATEGORY OF CITED DOCUMENTS cularly relevant if taken alone cularly relevant if combined with anoth ment of the same category nological background	T : theory or princi E : earlier patent d after the filing d er D : document cited L : document cited	ocumer ate in the a for othe	nt, but publis application er reasons	hed on, or



Application Number

EP 11 16 6330

	Citation of document with indi	sation where appropriate	Relevant	CLASSIFICATION OF THE
Category	of relevant passage		to claim	APPLICATION (IPC)
Х	DATABASE UniProt [On	ine]	1-5,7, 12,13,15	
	13 September 2005 (20		,_,	
	"SubName: Full=Mucin			
	protein (MASP), putat XP002661624,	:ive;",		
	retrieved from EBI ac	ccession no.		
	UNIPROT:Q4DU48			
	Database accession no	o. Q4DU48		
	* sequence *			
χ	ATKINS ET AL: "A foi	ur-strand coiled-coil	5,13	
	model for some insect	: fibrous proteins",		
	JOURNAL OF MOLECULAR	BIOLOGY, LONDON, GB,		
	vol. 24, no. 1, 28 February 1967 (196	57-02-28), pages		
	139-140, IN17, XP0240)15918,		
	ISSN: 0022-2836, DOI:			
	10.1016/0022-2836(67) [retrieved on 1967-02			
γ	* the whole document		$ _{1-4}$,	TECHNICAL FIELDS
			6-12,14,	SEARCHED (IPC)
			15	
Х	YU T-Y ET AL: "Confo	ormation transition of	5.13	
	silk fibroin",			
	CA,,	01 01) VD00202740		
γ	1 January 1996 (1996 * the whole document		$ _{1,2,4},$	
•	one whole accament		6-12,14,	
			15	
	•	-/		
		/		
	The present search report has been Place of search	•		Examiner
	Munich	Date of completion of the search	Chay	
		11 May 2012		vanne, Franz
	ATEGORY OF CITED DOCUMENTS	T : theory or principle E : earlier patent doc	ument, but publisl	
Y∶part	icularly relevant if taken alone icularly relevant if combined with another	after the filing date D : document cited in	the application	
A : tech	ument of the same category nnological background	L : document cited fo		
	-written disclosure rmediate document	& : member of the sa document	me patent family,	corresponding



Application Number

EP 11 16 6330

Category	Citation of document with i		opriate,		evant laim	CLASSIFIC APPLICATI	ATION OF THE
X	SILVA-ZACARIN E C N FORMATION MECHANISM SALIVARY GLANDS OF (HYMENOPTERA: APIDA J. BIOSCI., vol. 28, no. 6, 200 XP009108882, * the whole documer	M, ET AL.: " IS IN THE LAR APIS MELLIFE AE)", D3, pages 753	VAL RA	5,13		AITLIOAII	
Υ	WO 03/020916 A (UNI RANDOLPH V [US]; HA GATES) 13 March 200 * abstract * * page 1, lines 16- * page 5, lines 17- * page 6, line 11 - * page 8, lines 18-	AYASHI CHERYL 03 (2003-03-1 -20 * -27 * - page 7, lin	Ÿ [US]; 3)	1-4 6-12 15	2,14,		
Υ	W0 2004/016651 A (UMASON SIMON [GB]; F 26 February 2004 (2 * abstract * * page 1, lines 5-8 * page 4, line 13 * page 7, lines 17- * page 9, lines 16- * page 10, line 19	POUCHKINA NAT. 2004-02-26) 3 * - page 5, lin -24 * -29 *	ĀLIA [GB]) e 4 *	1-4 6-12 15	2,14,	TECHNIC, SEARCHE	AL FIELDS D (IPC)
X	DATABASE UniProt [0 13 September 2005 ("SubName: Full=Muci protein (MASP), put XP002675803, retrieved from EBI UNIPROT:Q4DVE5 Database accession * sequence *	[2005-09-13), n-associated ative;", accession no		1-5	,7, 13,15		
	The present search report has	been drawn up for all	claims	1			
	Place of search	Date of com	pletion of the search	\top		Examiner	
	Munich	11 Ma	y 2012		Cha	vanne, F	ranz
X : part Y : part docu A : tech O : non	ATEGORY OF CITED DOCUMENTS ioularly relevant if taken alone ioularly relevant if combined with anot unent of the same category inological background written disclosure rmediate document		T: theory or principle E: earlier patent doc after the filing dat D: document cited in L: document cited fo	cument, i e n the app or other r	ring the ir out publis olication easons	vention hed on, or	



Application Number EP 11 16 6330

	DOCUMENTS CONSID	ERED TO BE RELEVANT		
Category	Citation of document with in of relevant pass	ndication, where appropriate, ages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
Х	DATABASE EMBL [Onli	ne]	1-5,7, 12,13,15	
	15 February 1995 (1 "R.norvegicus mRNA antigen-related pro XP002675804, retrieved from EBI EM_R0:X83546 Database accession * sequence *	for leucocyte common tein (3941 bp).", accession no.	12,13,13	
х	DATABASE EMBL [Onli	ne]	1-5,13, 15	
		IIH_XGC_tropSki1 Xenopus one IMAGE:7872497 3', accession no.		TECHNICAL FIELDS SEARCHED (IPC)
х	DATABASE EMBL [Onli	ne]	1-5,7, 12,13,15	
	20 November 2000 (2 "Chlamydomonas reir helicase (Mut6) mRN XP002675806, retrieved from EBI EM_PL:AF305070 Database accession * sequence *	hardtii DEAH-box RNA IA, complete cds.", accession no.	12,13,13	
	The present search report has	been drawn up for all claims		
	Place of search	Date of completion of the search		Examiner
	Munich	11 May 2012	Cha	vanne, Franz
X : parti Y : parti docu A : tech O : non	ATEGORY OF CITED DOCUMENTS ioularly relevant if taken alone ioularly relevant if combined with anot ment of the same category nological background written disclosure mediate document	L : document cited for	ument, but publis the application rother reasons	hed on, or



Application Number EP 11 16 6330

ategory	Citation of document with indica of relevant passages	tion, where appropriate,	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
(DATABASE EMBL [Online]		1-5,13,	
	18 August 2000 (2000-0	08-18),	15	
	"WHE0517_A09_A17ZR Sec	cale cereale		
	aluminum-stressed root Secale cereale cDNA cl			
	mRNA sequence.",	OHE WHEOSI7_AUS_AI7,		
	XP002675807,			
	retrieved from EBI acc	cession no.		
	EM_EST:BE587138 Database accession no.	BF587138		
	* sequence *	22007 200		
	DATABASE Geneseq [Onli	nel	1-5,7,	
	· -	_	12,13,15	
	12 August 2004 (2004-0 "Human secreted protei	08-12), n SEO ID #3444 "		
	XP002675808,	Π 3LQ 1D #3444. ,		
	retrieved from EBI acc	cession no.		
	GSP:ADP31446 Database accession no.	ANP31446		TECHNICAL FIELDS
	* sequence *	7,0131110		SEARCHED (IPC)
(DATABASE Geneseq [Onli	nel	1-5,13,	
`	· -	_	15	
	29 August 2002 (2002-0			
	"Listeria monocytogene sequence #576.",	15 4D COILTY DINA		
	XP002675809,			
	retrieved from EBI acc GSN:ABQ70634	cession no.		
	Database accession no.	ABQ70634		
	* sequence *			
		-/		
		,		
	The present search report has been	drawn up for all claims		
	Place of search	Date of completion of the search		Examiner
	Munich	11 May 2012	Cha	vanne, Franz
C,	ATEGORY OF CITED DOCUMENTS	T : theory or principle E : earlier patent doc		
Y:part	cularly relevant if taken alone cularly relevant if combined with another	after the filing date D : document cited in	the application	•
docu A : tech	ment of the same category nological background	L : document cited for	r other reasons	
O : non	-written disclosure mediate document	& : member of the sar document	me patent family	, corresponding



Application Number

EP 11 16 6330

Category	Citation of document with in of relevant pass		ropriate,		elevant claim	CLASSIFICATION OF THE APPLICATION (IPC)
Х	DATABASE EMBL [Onli	ne]		1- 15	5,13,	
	2 July 2004 (2004-0	7-02).		13		
	"EST663692 RAB Rhip					
	appendiculatus cDNA		856 5' end,			
	mRNA sequence.",					
	XP002675810,	•				
	retrieved from EBI EM EST:CD792331	accession no).			
	Database accession	no CD792331				
	* sequence *	110. 60/ 52551	•			
	554.555					
X	DATABASE EMBL [Onli	ne]		1-	5,13,	
	30 September 2005 (
	"ppa01-30t3-e10 ppa					
	cDNA clone ppa01-30	it3-e10 5', n	IRNA			
	sequence.", XP002675811.					
	retrieved from EBI	accession no).			
	EM EST:DV099338		•			TECHNICAL FIELDS
	Database accession	no. DV099338	3			SEARCHED (IPC)
	* sequence *					
x	DATABASE EMBL [Onli	ne]			5,13,	
	27 August 2004 (200	M 00 27)		15		
	"WPAEhux1516F2 J12		lΩ ah1			
	Preamplified custom					
	pCMVsport6.1 (ResGe	n, Invitroge	en Inc.)			
	Emiliania huxleyi d	DNA, mRNA se	equence.",			
	XP002675812,	• • • • • •				
	retrieved from EBI	accession no).			
	EM_EST:CV070070 Database accession	no CV070076	1			
	* sequence *	110. 00070070	,			
			-/			
	The market by the state of the	do ()	Lalaina	1		
	The present search report has	'	npletion of the search	Ц,		Examiner
	Munich		•		Cha	vanne, Franz
		11 140	ay 2012			
	ATEGORY OF CITED DOCUMENTS		T : theory or principle E : earlier patent doo	umen		
Y∶part	icularly relevant if taken alone icularly relevant if combined with anot	her	after the filing dat D : document cited in	n the a		
docu	ıment of the same category nological background		L: document cited for	or othe	r reasons	
O : non	-written disclosure rmediate document		&: member of the same patent family, corresponding document			



Application Number EP 11 16 6330

	DOCUMENTS CONSID				
Category	Citation of document with in of relevant pass	ndication, where appropriate, ages		Relevant o claim	CLASSIFICATION OF THE APPLICATION (IPC)
Χ	DATABASE UniProt [0	Online]		5,7, ,13,15	
	13 September 2005 ("SubName: Full=Muciprotein (MASP), put XP002675813, retrieved from EBI UNIPROT:Q4E1Y4 Database accession * sequence *	n-associated surfactative;", accession no.		, - , - ,	
Х	DATABASE EMBL [Onli	ne]		5,13,	
	30 August 2001 (200 "BB170023B10C01.5 E Normalized/Subtract mellifera cDNA clor mRNA sequence.", XP002675814, retrieved from EBI EM_EST:BI508270 Database accession * sequence *	Bee Brain Led Library, BB17 A Led BB170023B10C01 5 Led accession no.		_	TECHNICAL FIELDS SEARCHED (IPC)
Х	DATABASE Geneseq [C	online]		5,7, ,13,15	
	3 June 2004 (2004-6 "S pneumoniae antig SeqID409.", XP002675815, retrieved from EBI GSP:ADM92212 Database accession * sequence *	genic protein seque		,10,10	
	The present search report has	been drawn up for all claims			
	Place of search	Date of completion of the	search		Examiner
	Munich	11 May 2012		Cha	vanne, Franz
X : part Y : part docu A : tech	ATEGORY OF CITED DOCUMENTS icularly relevant if taken alone icularly relevant if combined with anot iment of the same category nological background written disclosure	E : earlier p after the her D : docume L : docume	r principle under atent documer filing date and cited in the anticited for other same or of the	nt, but publisl application er reasons	hed on, or



Application Number

EP 11 16 6330

	DOCUMENTS CONSID	ERED TO BE RELEVAN	T		
Category	Citation of document with i of relevant pass	ndication, where appropriate, ages	Releva to claim		CLASSIFICATION OF THE APPLICATION (IPC)
Х	DATABASE EMBL [Onli	ne]	1-5,7, 12,13		
	5 October 1992 (1999) "Euprymna scolopes peroxidase-like process.", XP002675816, retrieved from EBI EM_INV:L01771 Database accession * sequence *	clone pLO7 otein mRNA, 3' end accession no.	12,13	, 13	
х	DATABASE EMBL [Onli	ne]	1-5,13 15	3,	
	fruit stored for 24	(ABDA) Royal Gala hours under low lus x domestica cDNA nRNA sequence.", accession no.			TECHNICAL FIELDS SEARCHED (IPC)
Х	DATABASE EMBL [Onli	ine]	1-5,7, 12,13		
	10 January 2003 (20 "Homo sapiens chron LA16c-448D5, WORKIN ordered piece.", XP002675819, retrieved from EBI EM_HTG:AC138510 Database accession * sequence *	nosome 16 clone NG DRAFT SEQUENCE, 1 accession no.			
	The present search report has	been drawn up for all claims			
	Place of search	Date of completion of the searc	ph		Examiner
	Munich	11 May 2012	(Chav	vanne, Franz
X : parti Y : parti docu A : tech O : non	ATEGORY OF CITED DOCUMENTS icularly relevant if taken alone icularly relevant if combined with anot iment of the same category nological background written disclosure mediate document	E : earlier pater after the filin her D : document ci L : document ci	inciple underlying nt document, but p g date ited in the applica ited for other reas	oublish tion ons	ned on, or



Application Number

EP 11 16 6330

CLAIMS INCURRING FEES
The present European patent application comprised at the time of filing claims for which payment was due.
Only part of the claims have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due and for those claims for which claims fees have been paid, namely claim(s):
No claims fees have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due.
LACK OF UNITY OF INVENTION
The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:
see sheet B
All further search fees have been paid within the fixed time limit. The present European search report has been drawn up for all claims.
As all searchable claims could be searched without effort justifying an additional fee, the Search Division did not invite payment of any additional fee.
Only part of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the inventions in respect of which search fees have been paid, namely claims:
None of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims, namely claims:
The present supplementary European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims (Rule 164 (1) EPC).



LACK OF UNITY OF INVENTION SHEET B

Application Number

EP 11 16 6330

The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

1. claims: 1-15(partially)

Polynucleotides of SEQ ID No.15, 16, 35 or 36 encoding silk proteins from honeybee or bumblebee, silk polypeptides comprising a sequence of SEQ ID No.5, 6, 26 or 27, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

2. claims: 1-15(partially)

Polynucleotides of SEQ ID No. 11, 12, 31 or 32 encoding silk proteins from honeybee or Bumblebee, polypeptides comprising a sequence of SEQ ID No.1, 2, 22 or 23, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

3. claims: 1-15(partially)

Polynucleotides of SEQ ID No.48, 49, 64 or 65 encoding silk polypeptides from bulldog ant and weaver ant, polypeptides comprising a sequence of SEQ ID No.40, 41, 56 or 57, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

4. claims: 1-15(partially)

Polynucleotides of SEQ ID No.13, 14, 33 or 34 encoding silk polypeptide from honeybee or bumblebee, polypeptides comprising a sequence of SEQ ID No.3, 4, 24 or 25, vector comprising said polynucleotide, host cell comprising said



LACK OF UNITY OF INVENTION SHEET B

Application Number

EP 11 16 6330

The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

5. claims: 1-15(partially)

Polynucleotides of SEQ ID No.50, 51, 66 or 67 encoding silk polypeptide from bulldog ant or weaver ant, polypeptides comprising a sequence of SEQ ID No.42, 43, 58 or 59, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

6. claims: 1-15(partially)

Polynucleotides of SEQ ID No.52, 53, 68 or 69 encoding silk polypeptides from bulldog ant or weaver ant, polypeptides comprising a sequence of SEQ ID No.44, 45, 60 or 61, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

7. claims: 1-15(partially)

Polynucleotides of SEQ ID No.17, 18, 37, 38 or 76 encoding silk polypeptides from honeybee or bumblebee, polypeptides comprising a sequence of SEQ ID No.7, 8, 28, 29 or 77, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said



LACK OF UNITY OF INVENTION SHEET B

Application Number

EP 11 16 6330

The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

polypeptide, kit comprising said polypeptide or polynucleotide.

8. claims: 1-15(partially)

Polynucleotides of SEQ ID No.54, 55, 70 or 71 encoding silk polypeptides from bulldog ant or weaver ant, polypeptides comprising a sequence of SEQ ID No.46, 47, 62 or 63, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

9. claims: 1-15(partially)

Polynucleotides of SEQ ID No.74 or 75 encoding silk polypeptides from lacewing, polypeptides comprising a sequence of SEQ ID No.72 or 73, vector comprising said polynucleotide, host cell comprising said vector, transgenic plant or non-human animal transfected with said polynucleotide, antibody specifically binding to said polypeptide, silk fiber comprising said polypeptide, copolymer comprising said polypeptide, composition comprising said polypeptide or said polynucleotide, method of treatment based on the use of said polypeptide, kit comprising said polypeptide or polynucleotide.

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO.

EP 11 16 6330

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

11-05-2012

c	Patent document ited in search report		Publication date		Patent family member(s)	Publication date
W	0 03020916	A	13-03-2003	AU CA EP JP WO	2002306944 A1 2459777 A1 1427742 A1 2005502347 A 03020916 A2	18-03-200 13-03-200 16-06-200 27-01-200 13-03-200
- W	0 2004016651	Α	26-02-2004	AU WO	2003260719 A1 2004016651 A2	03-03-200 26-02-200
-						
			ficial Journal of the Eurc			